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REQUEST FOR CERTIFICATE OF CORRECTION UNDER 37 CFR 1.322
Docket No. MA-43CDF2D3
Patent No. 6,737,273

Jay M. Sanders, Patent Attorney

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants : Jewel Payne, August J. Sick
Issued : May 18, 2004
Patent No. : 6,737,273 *b2*
For : *Bacillus thuringiensis* Isolate Active Against Lepidopteran Pests, and Genes Encoding Novel Lepidopteran-Active Toxins

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Certificate
AUG 08 2004
of Correction

REQUEST FOR CERTIFICATE OF CORRECTION
UNDER 37 CFR 1.322 (OFFICE MISTAKE)

Sir:

A Certificate of Correction for the above-identified patent has been prepared and is attached hereto.

In the left-hand column below is the column and line number where errors occurred in the patent. In the right-hand column is the page and line number in the application where the correct information appears.

Patent Reads:

Column 1, Line 56:
“toxin gene PS81I2”

Column 3, Line 51:
“then can be If applied”

Column 6, Line37:
“Nitrobacteraceae, Among”

Application Reads:

Page 2, Line 12:
--toxin gene PS81IA2--

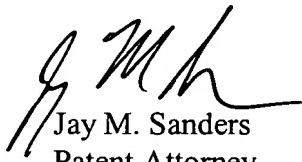
Page 5, Line 12-13:
--then can be applied--

Page 9, Line 37:
-- Nitrobacteraceae. Among--

A true and correct copy of pages 2, 5, and 9 of the specification as filed which supports Applicant's assertion of the error on the part of the Patent Office accompanies this Certificate of Correction.

Approval of the Certificate of Correction is respectfully requested.

Respectfully submitted,



Jay M. Sanders

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Attachments: Certificate of Correction; copy of pages 2, 5, and 9 of the specification

UNITED STATES PATENT AND TRADEMARK OFFICE

CERTIFICATE OF CORRECTION

PATENT NO. : 6,737,273 *B2*

DATED : May 18, 2004

INVENTORS : Jewel Payne, August J. Sick

It is certified that errors appear in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

Column 1:

Line 56, "toxin gene PS81I2" should read --toxin gene PS81IA2--

Column 3:

Line 51, "then can be If applied" should read --then can be applied—

Column 6:

Line37, "Nitrobacteraceae, Among" should read --Nitrobacteraceae. Among--

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PATENT NO. 6,737,273 *B2*

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FORM PTO-1050 (REV. 3-75) UNITED STATES PATENT AND TRADEMARK OFFICE

AUG 09 2004

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FORM PTO-1050 (REV. 3-75) UNITED STATES PATENT AND TRADEMARK OFFICE

AUG 09 2004



Brief Summary of the Invention

The subject invention concerns a novel Bacillus thuringiensis isolate designated B.t. PS81I which has activity against all lepidopteran pests tested.

Also disclosed and claimed are novel toxin genes which express toxins toxic to 5 lepidopteran insects. These toxin genes can be transferred to suitable hosts via a plasmid vector.

Specifically, the invention comprises the novel B.t. isolate denoted B.t. PS81I, mutants thereof, and novel δ-endotoxin genes derived from this B.t. isolate which encode proteins which are active against lepidopteran pests.

10

Brief Description of the Sequences

SEQ ID NO:1 is the nucleotide sequence of the novel *B.t.* toxin gene PS81IA2.

SEQ ID NO:2 is the amino acid sequence of the novel *B.t.* toxin PS81IA2.

SEQ ID NO:3 is the nucleotide sequence of the novel *B.t.* toxin gene PS81B.

15

SEQ ID NO:4 is the amino acid sequence of the novel *B.t.* toxin PS81B.

SEQ ID NO:5 is the nucleotide sequence of the novel *B.t.* toxin gene PS81IB2.

SEQ ID NO:6 is the amino acid sequence of the novel *B.t.* toxin PS81IB2.

SEQ ID NO:7 is the nucleotide sequence of the novel *B.t.* toxin gene PS81IA.

SEQ ID NO:8 is the amino acid sequence of the novel *B.t.* toxin PS81IA.

20

Brief Description of the Drawings

Figure 1 – agarose gel electrophoresis of plasmid preparations from B.t. HD-1 and B.t. PS81I.

25

Detailed Disclosure of the Invention

The novel toxin genes of the subject invention were obtained from a novel lepidopteran-active B. thuringiensis (B.t.) isolate designated PS81I.

or for the enforceable life of any patent which may issue disclosing the cultures. The depositor acknowledges the duty to replace the deposits should the depository be unable to furnish a sample when requested, due to the condition of the deposit(s). All restrictions on the availability to the public of the subject culture deposits will be irrevocably removed upon
5 the granting of a patent disclosing them.

The toxin genes of the subject invention can be introduced into a wide variety of microbial hosts. Expression of the toxin gene results, directly or indirectly, in the intracellular production and maintenance of the pesticide. With suitable hosts, e.g.,
10 Pseudomonas, the microbes can be applied to the situs of lepidopteran insects where they will proliferate and be ingested by the insects. The result is a control of the unwanted insects. Alternatively, the microbe hosting the toxin gene can be treated under conditions that prolong the activity of the toxin produced in the cell. The treated cell then can be applied to the environment of target pest(s). The resulting product retains the toxicity of the
B.t. toxin.
15

Where the B.t. toxin gene is introduced via a suitable vector into a microbial host, and said host is applied to the environment in a living state, it is essential that certain host microbes be used. Microorganism hosts are selected which are known to occupy the "phytosphere" (phyloplane, phyllosphere, rhizosphere, and/or rhizoplane) of one or more crops of interest. These microorganisms are selected so as to be capable of successfully
20 competing in the particular environment (crop and other insect habitats) with the wild-type microorganisms, provide for stable maintenance and expression of the gene expressing the polypeptide pesticide, and, desirably, provide for improved protection of the pesticide from environmental degradation and inactivation.

A large number of microorganisms are known to inhabit the phylloplane (the surface of the plant leaves) and/or the rhizosphere (the soil surrounding plant roots) of a wide variety of important crops. These microorganisms include bacteria, algae, and fungi. Of particular interest are microorganisms, such as bacteria, e.g., genera Bacillus, Pseudomonas, Erwinia, Serratia, Klebsiella, Xanthomonas, Streptomyces, Rhizobium, Rhodopseudomonas, Methylophilus, Agrobacterium, Acetobacter, Lactobacillus, Arthrobacter, Azotobacter,

which will include at least one replication system, but may include more than one, where one replication system is employed for cloning during the development of the plasmid and the second replication system is necessary for functioning in the ultimate host. In addition, one or more markers may be present, which have been described previously. Where integration
5 is desired, the plasmid will desirably include a sequence homologous with the host genome.

The transformants can be isolated in accordance with conventional ways, usually employing a selection technique, which allows for selection of the desired organism as against unmodified organisms or transferring organisms, when present. The transformants then can be tested for pesticidal activity.

10 Suitable host cells, where the pesticide-containing cells will be treated to prolong the activity of the toxin in the cell when the then treated cell is applied to the environment of target pest(s), may include either prokaryotes or eukaryotes, normally being limited to those cells which do not produce substances toxic to higher organisms, such as mammals. However, organisms which produce substances toxic to higher organisms could be used,
15 where the toxin is unstable or the level of application sufficiently low as to avoid any possibility of toxicity to a mammalian host. As hosts, of particular interest will be the prokaryotes and the lower eukaryotes, such as fungi. Illustrative prokaryotes, both Gram-negative and -positive, include Enterobacteriaceae, such as Escherichia, Erwinia, Shigella, Salmonella, and Proteus; Bacillaceae; Rhizobaceae, such as Rhizobium; Spirillaceae, such as photobacterium, Zymomonas, Serratia, Aeromonas, Vibrio, Desulfovibrio, Spirillum; Lactobacillaceae; Pseudomonadaceae, such as Pseudomonas and Acetobacter; Azotobacteraceae, Actinomycetales, and Nitrobacteraceae. Among eukaryotes are fungi, such as Phycomycetes and Ascomycetes, which includes yeast, such as Saccharomyces and Schizosaccharomyces; and Basidiomycetes yeast, such as Rhodotorula, Aureobasidium,
20 Sporobolomyces, and the like.
25

Characteristics of particular interest in selecting a host cell for purposes of production include ease of introducing the B.t. gene into the host, availability of expression systems, efficiency of expression, stability of the pesticide in the host, and the presence of auxiliary genetic capabilities. Characteristics of interest for use as a pesticide microcapsule include